bio-protocol Preprint

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Sperm motility

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An abbreviated version of this protocol was published in eLIFE in Dec 2015

A K^+-selective CNG channel orchestrates Ca^{2+} signalling in zebrafish sperm

DOI: 10.7554/eLife.07624

Detailed protocol

The NP-EGTA-AM was diluted in DMSO at 4 mM stock solutions. We recommend using water-free ampullas of DMSO.

The incubation protocol was:
- 94 µl sample (in ES solution)
- 5 µl Pluronic 10% (0.05% Final concentration)
- 1 µl NP-EGTA AM Caged Calcium 4 mM (40 µM Final concentration)

Incubation was performed at room temperature (about 22°C in our lab) for 45 - 60 minutes. 60 minutes incubations sometimes resulted in better responses.
We diluted the incubation 1:20 in activation solution. At this point, you have to hurry, as cells are only motile for a short period of time. If cells are sticking to the walls of the observation chamber, you can add 0.05% final pluronic in your final sperm suspension.
NP-EGTA is a poor caged compound. Therefore, use as much light for uncaging as possible.

How to cite:(Readers should cite both the Bio-protocol preprint and the original research article where this protocol was used)

1. Fechner, S. , Kaupp, U. and Seifert, R. (2021). Sperm motility. Bio-protocol Preprint. bio-protocol.org/prep812.
2. Fechner, S., Alvarez, L., Bönigk, W., Müller, A., Berger, T. K., Pascal, R., Trötschel, C., Poetsch, A., Stölting, G., Siegfried, K. R., Kremmer, E., Seifert, R. and Kaupp, U. B.(2015). A K^+-selective CNG channel orchestrates Ca^{2+} signalling in zebrafish sperm. eLIFE. DOI: 10.7554/eLife.07624

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